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Spring 2004 Sudden Oak Death Detection Survey in Washington State

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Summary

Phytophthora ramorum, the causal agent of "sudden oak death" (SOD), is responsible for widespread tree mortality in central and northern California. Western Washington is at high risk for SOD due to the presence of known SOD hosts in the natural environment, suitable climatic conditions (extended periods of moist weather and mild temperatures), and the presence of nurseries receiving known SOD host stock. Between March 2003 and March 2004, ninety Washington nurseries received stock from a nursery in California that contained and distributed host stock, which later tested positive for P. ramorum. Between March 24, 2004 and June 1, 2004, Washington DNR surveyed 42 nurseries (159 transects) and 7 general forest sites (27 transects). One hundred and forty-four foliage samples were collected from known (and potential) SOD hosts including Rhododendron spp., bigleaf maple, Douglas-fir, evergreen huckleberry and Pacific madrone. The samples were screened for Phytophthora spp. by the Washington State Department of Agriculture using DAS (Double Antibody Sandwich) ELISA kits. Twelve samples tested positive for Phytophthora spp. and were sent to Oregon State University to be tested for P. ramorum using PCR (Polymerase Chain Reaction) based diagnostic tests. All of the samples tested negative for P. ramorum.

Introduction

Phytophthora ramorum, the cause of "sudden oak death (SOD)," is responsible for widespread tree mortality in central and northern California. Since its discovery in 1995 on tanoak in Marin County, California, SOD has spread to 14 counties in California and a small area near the town of Brookings, Oregon. In 2003, the disease was found in nurseries in Washington, Oregon, and British Columbia, Canada.

In 2003, the USDA Forest Service completed a risk assessment pertaining to SOD. Based on the number of susceptible hosts, climatic conditions and locations of nurseries receiving SOD host

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stock, western Washington is considered to be at high risk for this disease. In addition, shipments of potentially infected stock from a nursery in California that tested positive for *P. ramorum* increased the risk of infection in the perimeters of the 90 nurseries that received the stock. The objectives of this survey were to assess the perimeters of nurseries that had received stock from the infected nursery in California and to gather information on the distribution of SOD in the high-risk zone of Washington.

Sudden Oak Death received its name because of the devastation it has caused on tanoaks, coast live oaks, and other members of the black oak family (*Erythrobalanus*) in California. At this point in time, it is not known to affect members of the white oak family (*Lepidobalanus*), including Oregon white oak, which is the only oak native to Washington. The current list of susceptible (regulated) host species that are native to western Washington includes rhododendron, Douglas-fir, bigleaf maple, Pacific madrone, evergreen huckleberry, wood rose, western starflower and California honeysuckle. Other "associated hosts" (found naturally infected, but Koch's postulates have not been completed) native to Washington include grand fir, cascara, salmonberry, false Solomon's seal, Pacific yew, poison oak, California hazelnut and California wood fern. Although these species are the only western Washington hosts currently recognized, new host species are being identified as susceptibility trials continue.

Biology

Phytophthora ramorum is a fungus-like organism that can infect a wide range of host species. The organism produces microscopic reproductive, dispersal and survival structures making definitive field identification of the disease impossible. Sporangia are the reproductive structures of the organism and are known to play a significant role in spreading the disease. Sporangia may germinate and directly infect plants, but more importantly, they can produce and release zoospores. Zoospores are motile spores (spores with two flagella) that swim in free water and infect plants through wounds or succulent plant tissue. Sporangia are often found on the foliage of hosts and are generally spread by rain splash, or aerially, to new hosts. Chlamydospores are survival structures that can endure months of adverse conditions and germinate when suitable conditions are present. All three structures require free water for germination. Chlamydospores are often present in the soil surrounding infected plants and can be transported with soil on shoes, tires, animal paws/hooves, etc. aiding in long-range dispersal of the organism. Spores can also be transported in stream water.

Symptoms

Symptoms caused by *P. ramorum* vary among host species. *Phytophthora ramorum* primarily infects the leaves of *Rhododendron* spp., although it can also infect branches and may kill entire plants. Infection causes brown-black (necrotic) lesions on portions of the leaf often where water accumulates. The lesions have a diffuse margin and can mimic drought injury. *Phytophthora ramorum* can cause similar symptoms on Pacific madrone and may also cause leaf spots, necrosis along the midvein of the leaf, leaf death, and branch dieback. Bigleaf maple is a foliar host of *P. ramorum* and symptoms tend to look like scorch starting at the edge of the leaf and

with irregular margins that do not follow the contour of the leaf. Occasional leaf spots have been identified on evergreen huckleberry, but branch dieback is a more common symptom. *Phytophthora ramorum* causes small lesions on twigs or stems of evergreen huckleberry resulting in death of the infected branch or stem beyond the lesion. *Phytophthora ramorum* also causes cankers on small branches of Douglas-fir resulting in tip dieback, which looks very similar to frost damage. It is important to note that abiotic stressors such as drought and frost, as well as other biological agents, can produce symptoms similar to those caused by *P. ramorum* in the above hosts. Only laboratory analysis of symptomatic tissue can confirm the presence of *P. ramorum*.

Methods

The sampling protocol utilized in this survey was developed by the Forest Health Monitoring group within the USDA Forest Service. In forested areas around nurseries that contained suitable host types, four 100-m long transects were distributed along the nursery perimeter so as to sample all available aspects (Fig. 1). The transects encompassed the edge of the forested area along the nursery perimeter and extended 10 meters into the forested area (Fig. 1).

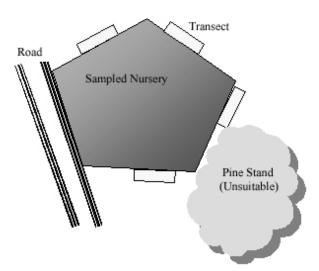


Figure 1. Sampling protocol for nursery perimeter surveys.

In high-risk forest environments, not adjacent to nurseries, a plot center was located at least 100 meters from a road. Four 100-m transects, each 10-m wide, were installed on cardinal azimuths from the plot center. A Global Positioning System (GPS) was used to identify starting and ending coordinates for each transect.

Along each transect rhododendron, evergreen huckleberry, bigleaf maple, Douglas-fir, Pacific madrone and other known and potential hosts were examined for symptoms of SOD. Samples of species not currently recognized as hosts were collected if they exhibited symptoms similar to those exhibited by known hosts because of the likelihood that there are unidentified hosts that

will be listed as susceptible in the future. Symptomatic leaves were placed in labeled plastic bags and delivered to the laboratory, usually within the same day. The samples were tested by the Washington State Department of Agriculture using DAS (Double Antibody Sandwich) ELISA kits. Samples that tested positive for *Phytophthora* spp. using the ELISA kits were sent to Oregon State University to be tested for *P. ramorum* using PCR (Polymerase Chain Reaction) based diagnostic tests.

Results and Discussion

The forested perimeter of 42 nurseries, including 159 transects with suitable hosts, were surveyed for SOD (Fig. 2). The absence of a suitable forested perimeter limited the number of transects completed on some nurseries. Twenty-seven transects in seven forested areas were surveyed for SOD (Fig 2.).

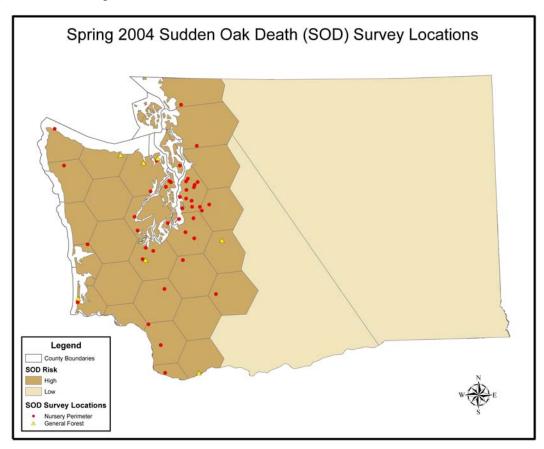


Figure 2. Location of spring 2004 nursery perimeter and general forest SOD survey sites within USDA Forest Service SOD risk polygons.

Phytophthora ramorum was not isolated from any of the 144 samples collected (Table 1). *Rhododendron* spp. were the most common species sampled, followed by bigleaf maple and vine maple (Table 1).

Table 1. Samples collected from nursery perimeter and general forest transects during the spring 2004 SOD survey and the results of the *Phytophthora ramorum* analysis.

Species	Number of Samples - Nursery Perimeter	Number of Samples - General Forest	Total Number of Samples	P. ramorum +/-
Rhododendron spp.	53	9	62	=
Bigleaf maple	14	0	14	-
Vine maple	7	4	11	-
Japanese maple	5	0	5	-
No. red oak	5	0	5	-
Red maple	4	1	5	-
Madrone	4	0	4	-
Viburnum spp.	3	0	3	-
Salal	3	0	3	-
Douglas-fir	2	0	2	-
Coast redwood	2	0	2	-
Vaccinium spp.	2	0	2	=
Pieris spp.	2	0	2	-
Wild rose	2	1	3	-
Quercus spp.	2	0	2	-
Oregon grape	0	2	2	-
Evergreen huckleberry	1	0	1	-
Red huckleberry	1	0	1	1
Grand fir	1	0	1	1
Photinia spp.	1	0	1	-
English laurel	1	0	1	-
Norway maple	1	0	1	ı
Oregon ash	1	0	1	ı
Malus spp.	1	0	1	-
Prunus spp.	1	0	1	-
Indian plum	1	0	1	-
Magnolia spp.	1	0	1	-
Aucuba japonica	1	0	1	-
Unknown host	5	0	5	-
Totals	127	17	144	

While SOD has now been found in 25 nurseries in Washington, there is no evidence that the disease has spread beyond the confines of those nurseries. Infected plants within the nurseries were destroyed to prevent further spread of the pathogen. The organism was brought to the Washington nurseries on plants received from infected nurseries in California and Oregon. There is no evidence that the organism has spread naturally in the environment from California, or Oregon, to Washington.